

THE POLLINATION ECOLOGY  
OF FIVE SPECIES OF  
*PENSTEMON*

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(SCROPHULARIACEAE) IN  
THE TALLGRASS PRAIRIE<sup>1</sup>

ABSTRACT

The floral ecology of *Penstemon cobaea* Nutt. var. *cobaea*, *P. cobaea* var. *purpureus* Pennell, *P. digitalis* Nutt. ex Sims, *P. grandiflorus* Nutt., *P. pallidus* Small, and *P. tubaeformis* Nutt. was studied by sampling populations at nine prairie sites in Illinois, Kansas, and Missouri. All five species show protandry, but the receptive stigma lies only 2 mm away from the two pairs of fertile stamens. Bagging experiments on three *Penstemon* spp. show that only *P. digitalis* sets seed when insect visitors are excluded. Flowers of all five species exhibit a horizontal presentation of the corolla and emit a slightly discernible scent. However, the presumed pollination systems of *Penstemon* spp. correlate with corolla form and the size of the floral sinns. The tubular, white flowers of *P. tubaeformis* appear to be pollinated by a combination of diurnal Lepidoptera and some native bees favoring a dorsal deposition of pollen on mouthparts and upper thoraces. The four remaining species have gullet- or bell-shaped corollas ornamented with violet-purple blotches or lines. These species appear to be pollinated primarily by polylectic/polyphagous bees (including six *Bombus* spp.), with *P. digitalis* and *P. cobaea* visited infrequently by the rare *Penstemon* wasp, *Pseudomasaris occidentalis*. Queens of *Bombus pennsylvanicus* subsp. *pennsylvanicus* forage primarily on the large, gullet flowers of *P. grandiflorus* and both varieties of *P. cobaea*. Queens of *Bombus nevadensis* subsp. *auricomus* prefer those *P. digitalis* and *P. pallidus* showing a reduced gullet or bell form. Bee pollination in four *Penstemon* spp. may operate within a two-tiered system. Large-bodied *Bombus* spp., *Megachile brevis*, and anthophorids (*Synhalonia hamata* and *Anthophora terminalis*) carry dorsal depositions of *Penstemon* pollen because they contact anthers and stigmas while they forage exclusively for nectar. In contrast, small-bodied members of the Anthophoridae (*Ceratina*), Colletidae (*Hylaeus* spp.), Halictidae (*Augochlorella*, *Halictus*, *Lasiglossum*), and Megachilidae (*Hoplitis* and *Osmia*) forage actively for *Penstemon* pollen encouraging repeated, ventral contact with the sexual organs of the flowers. *Bombus* queens were more prevalent at large *Penstemon* populations (especially *P. cobaea*). *Bombus* workers were collected primarily on restored sites. The importance of small bees as pollinators appeared to vary indirectly with *Penstemon* population size.

*Penstemon* (Scrophulariaceae: Cheloneae) is a North American genus of about 270 species (Wolfe et al., 1997) distributed from Alaska to Guatemala. Within the Great Plains, Freeman (1981) recognized 22 species of *Penstemon* in two subgenera and five sections. The Upper Mississippi Valley supports two additional species, *Penstemon arkansanus* Pennell and *P. hirsutus* (L.) Willd., suggesting that 24 *Penstemon* species are native to midwestern American prairies.

Despite the species richness of *Penstemon* in North America, analyses comparing life-histories within this genus lag far behind classical (Pennell, 1935; Keck, 1938) and molecular (Wolfe & Elisens, 1993) taxonomies. We lack significant literature on

breeding systems in *Penstemon* compared to other scrophulariaceous genera (e.g., *Pedicularis*; Macior, 1982) distributed through the Northern Hemisphere (Kampry, 1995). References to the pollination biology of *Penstemon* by Pennell (1935) were derived primarily from predictions that were based on floral morphology. In contrast, what literature does exist on *Penstemon* pollination often shows a lack of consensus regarding the efficiency of different bee taxa as true pollen vectors.

Field studies of *Penstemon* pollination began with Robertson (1892, 1929), who noted protandry in "*P. laevigatus*" (= *P. digitalis*) and "*P. pubescens*" (= *P. pallidus*) in Illinois and collected a total of 20 different bee species in their flowers. Rob-

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erson was convinced that pollination was effected exclusively by long-tongued bees probing for nectar because small-bodied, short-tongued bees were excluded from the floral throat by the ornamented staminode. Clements and Long (1923) collected floral foragers on *P. gracilis*, in the Colorado foothills, concluding that *Osmia* and *Hoplitis* spp. (Megachilidae) were the most common floral visitors. Straw (1956) insisted that the pseudomasarid wasp-pollinated *Penstemon spectabilis* Thurb. was derived from a stabilized hybrid between bee-pollinated *P. grinnellii* Eastw. and hummingbird-pollinated *P. centrathifolius* (Benth.) Benth. This has since been discredited by Grant (1994; see also Wolfe & Elisens, 1993). Crosswhite and Crosswhite (1966) studied insect-pollination of *P. gracilis* and *P. pallidus* in southern Wisconsin and northern Illinois. Unlike Robertson, they emphasized the role of small, solitary bees, especially *Osmia* and *Hoplitis* spp. Crosswhite (1965) provided the only information on compatibility systems in the genus, concluding that *P. pallidus* was self-compatible. Lawson et al. (1989) analyzed the pollen loads of bees collected on *Penstemon haydenii* S. Wats. in Nebraska, noting an abundance of *Hoplitis* and *Osmia*, but did not comment on which bees regularly contacted the receptive stigmas.

Here we report the results of four years of field studies on six *Penstemon* taxa in five species: *Penstemon cobaea* var. *cobaea*, *P. cobaea* var. *purpureus*, *P. digitalis*, *P. grandiflorus*, *P. pallidus*, and *P. tubaeiflorus*. Continued studies of pollination systems of this genus in the American Midwest are needed for two, overlapping reasons. First, further analyses and descriptions of pollination mechanisms in *Penstemon* will make it possible to map pollinator shifts onto a phylogenetic tree (see Armbruster, 1993; Goldblatt et al., 1995). Second, *Penstemon* spp. are native to prairies that are reduced and much endangered habitats through the American Midwest. Basic information on pollen dispersal in prairie *Penstemon* spp. should contribute ultimately to conservation policies and restoration projects.

#### MATERIALS AND METHODS

The taxonomy of *Penstemon* species follows Yatskevych and Turner (1990). Floral phenologies are based on censuses (as specified below) at each study site for one to four flowering seasons.

#### POPULATIONS

*Penstemon cobaea* Nutt. var. *cobaea* (pollinators collected 24 May–22 June 1994–1997,  $N = 116$ ). Sixteen populations of this taxon on or near the

Konza Prairie (Kansas) have been studied, most of them small ( $< 30$  blooming stems). The Konza Prairie Research Natural Area is an 8616-acre preserve managed by Kansas State University for the Nature Conservancy. Two large populations ( $> 500$  blooming stems) were studied: (1) in 1995 in White Pasture near the northeastern corner of the Konza in an area considered more floristically rich in tall-grass prairie forbs than much of the rest of the site; and (2) in Wright Prairie, adjacent to the part of Konza known as the Texas Hog Pasture, for which access was provided by Valerie Wright.

*Penstemon cobaea* var. *purpureus* Pennell (28 May–4 June 1995–1996,  $N = 15$ ). Three populations of approximately 100 flowering shoots each were monitored in roadside glades in Christian, Ozark, and Taney Cos., Missouri. All three sites are listed on the Missouri Department of Conservation Rare Plant Inventory.

*Penstemon digitalis* Nutt. ex Sims (30 May–5 July 1994–1996,  $N = 325$ ). This species was studied on four sites, two tallgrass prairie restorations in eastern Missouri and two tallgrass prairie relicts in western Missouri: (1) Litzinger Road Ecology Center of the Missouri Botanical Garden, St. Louis Co., Missouri. This wet mesic prairie restoration was seeded in 1989 and still contains a preponderance of weedy, early successional prairie species. (2) Shaw Arboretum Experimental Prairie of the Missouri Botanical Garden, Franklin Co., Missouri. This site is about 25 years old and is more diverse than Litzinger, but not so diverse as the natural communities. (3) Paint Brush Prairie, Pettis Co., Missouri. The colony studied is in a relatively impoverished part of the site, but is adjacent to floristically rich prairie. There are notable differences, discussed below, between the floral foragers here as compared to the restorations. (4) Hi-Lonesome Prairie, Benton Co., Missouri. The study area here is a *Penstemon* monoculture embedded in common lenticular sedges (*Carex* spp.).

*Penstemon grandiflorus* Nutt. (21–30 May 1996–1997,  $N = 44$ ). This species was studied in a population of about 50 blooming plants on and near an ungrazed prairie haymeadow on the Poole Ranch in Geary Co., Kansas, about 2.8 miles south of Interstate 70 on Kansas Hwy. 177. This species is extremely rare on the Konza Prairie proper.

*Penstemon pallidus* Small (15–24 May 1996–1997,  $N = 71$ ). Two sites were used for this taxon in both 1996 and 1997: (1) Fults Hill Prairie, Monroe Co., Illinois. This area is perched atop the Mississippi River bluffs and is considered the best surviving undisturbed loess hill prairie along the Illinois bluffs. (2) Paint Brush Prairie, Pettis Co.,

Table 1. Pollen loads of foragers collected on *Penstemon* spp.

	Penstemon pollen only	Penstemon + other species	Other species only	No pollen	Total
<b>Large Gullet-Corolla Species</b>					
<b><i>Penstemon cobaea</i> var. <i>cobaea</i></b>					
<b>Bees</b>					
<i>Augochlorella striata</i> (Provancher) F	2	0	0	0	2
<i>Bombus nevadensis auricomus</i> (Robt.) Q	4	1	0	0	5
<i>Bombus nevadensis auricomus</i> W	2	0	0	0	2
<i>Bombus pennsylvanicus</i> (DeGeer) Q	19	8	2	1	30
<i>Bombus pennsylvanicus</i> W	1	0	0	0	1
<i>Ceratina strenua</i> (Smith) F	0	1	0	0	1
<i>Halictus ligatus</i> Say F	1	0	0	0	1
<i>Hoplitis pilosifrons</i> (Cresson) F	21	21	3	5	50
<i>Hoplitis pilosifrons</i> M	0	0	0	1	1
<i>Lasioglossum</i> (= <i>Dialictus</i> ) spp. F	6	2	0	1	9
<i>Synhalonia hamata</i> (Bradley) F	1	0	0	0	1
<b>Others</b>					
<i>Bombylius</i> sp.	4	0	0	3	7
<i>Euphoria sepulchralis</i> (Fab.)	0	2	0	4	6
					Σ = 116
<b><i>Penstemon cobaea</i> var. <i>purpureus</i></b>					
<b>Bees</b>					
<i>Bombus pennsylvanicus</i> Q	0	0	0	1	1
<i>Hoplitis pilosifrons</i> F	1	2	0	1	4
<i>Lasioglossum imitatum</i> (Smith) F	1	0	0	0	1
<i>Osmia distincta</i> (Cresson) F	1	1	0	0	2
<i>Synhalonia rosae</i> Robertson F	1	0	0	0	1
<i>Xylocopa virginica</i> L. F	0	1	1	1	3
<b>Others</b>					
<i>Bombylius</i> sp.	0	0	0	1	1
<i>Pseudomasaris</i> sp. F	1	0	0	1	2
					Σ = 15
<b><i>Penstemon grandiflorus</i></b>					
<b>Bees</b>					
<i>Augochlorella persimilis</i> (Viereck) F	1	0	0	0	1
<i>Augochlorella striata</i> F	11	2	1	4	18
<i>Bombus griseocolitis</i> (DeGeer) Q	0	1	0	0	1
<i>Bombus pennsylvanicus</i> Q	5	3	0	0	8
<i>Hoplitis pilosifrons</i> F	3	0	0	0	3
<i>Lasioglossum</i> (= <i>Dialictus</i> ) spp. F	6	7	0	1	14
<i>Megachile brevis</i> Say M	1	0	0	0	1
<i>Xylocopa virginica</i> M	1	0	0	0	1
					Σ = 44
<b>Small Gullet-Corolla Species</b>					
<b><i>Penstemon digitalis</i></b>					
<b>Bees</b>					
<i>Anthophora terminalis</i> Cresson F	6	0	0	1	7
<i>Augochlorella striata</i> F	3	1	0	3	7
<i>Bombus bimaculatus</i> Cresson Q	2	0	1	2	5
<i>Bombus bimaculatus</i> W	24	8	6	3	41
<i>Bombus bimaculatus</i> M	5	0	0	0	5
<i>Bombus fraternus</i> (Smith) W	8	1	1	0	10

Table 1. Continued.

	Penstemon pollen only	Penstemon + other species	Other species only	No pollen	Total
<i>Bombus griseocollis</i> Q	0	1	1	2	4
<i>Bombus griseocollis</i> W	3	5	0	1	9
<i>Bombus impatiens</i> Cresson W	9	1	1	0	11
<i>Bombus nevadensis</i> Q	28	2	6	9	45
<i>Bombus nevadensis</i> W	5	5	4	2	16
<i>Bombus pennsylvanicus</i> Q	3	3	1	2	9
<i>Bombus pennsylvanicus</i> W	7	6	1	2	16
<i>Ceratina</i> spp. F	7	5	0	5	17
<i>Hoplitis pilosifrons</i> F	6	2	0	1	9
<i>Hylaeus</i> spp. F	2	0	0	2	4
<i>Lasioglossum</i> (= <i>Dialictus</i> ) spp. F	36	5	3	12	56
<i>Osmia</i> spp. F	11	0	0	1	12
<i>Synhalonia hamata</i> F	22	13	2	2	39
<i>Synhalonia hamata</i> M	1	0	0	0	1
<b>Others</b>					
<i>Pseudomasaris</i> sp. F	1	0	0	0	1
<i>Pterourus troilus troilus</i> L. F	1	0	0	0	1
					Σ = 325
<b><i>Penstemon pallidus</i></b>					
<b>Bees</b>					
<i>Apis mellifera</i> L. W	1	0	0	0	1
<i>Augochlorella striata</i> F	0	0	1	1	2
<i>Bombus bimaculatus</i> Q	2	0	0	1	3
<i>Bombus bimaculatus</i> W	1	1	0	0	2
<i>Bombus nevadensis auricomus</i> Q	0	0	0	2	2
<i>Bombus pennsylvanicus</i> Q	0	0	0	2	2
<i>Ceratina</i> spp. F	2	0	0	5	7
<i>Hoplitis pilosifrons</i> F	13	1	0	6	20
<i>Hoplitis producta</i> (Cresson) F	1	0	0	0	1
<i>Lasioglossum</i> (= <i>Dialictus</i> ) sp. F	1	1	0	0	2
<i>Osmia</i> spp. F	15	3	0	6	24
<i>Synhalonia rosae</i> M	3	0	0	2	5
					Σ = 71
<b>Tubular-Corolla species</b>					
<b><i>Penstemon tubaeformis</i></b>					
<b>Bees</b>					
<i>Anthophora abrupta</i> Say F	0	0	0	2	2
<i>Anthophora abrupta</i> M	1	0	0	2	3
<i>Anthophora ursina</i> Cresson F	1	0	0	0	1
<i>Augochlorella striata</i> F	0	0	0	1	1
<i>Bombus pennsylvanicus</i> Q	3	0	0	0	3
<i>Osmia distincta</i> F	2	0	0	0	2
<b>Others</b>					
<i>Pterourus troilus troilus</i> F	1	0	0	2	3
<i>Pterourus troilus troilus</i> M	0	0	0	1	1
					Σ = 16
					ΣΣ = 587

F = Female, M = male, Q = queen, W = worker.

Missouri. This Osage Plains prairie is particularly rich in late spring forbs which co-flower with penstemons.

*Penstemon tubaeformis* Nutt. (4–5 June 1996,  $N = 17$ ). This species was studied at Long Bald, a dolomite glade in Caney Mountain Conservation Area, Ozark Co., Missouri. This is a large and spectacular glade surrounded by dry oak woods, and it contains a wealth of wildflowers, including the Ozark endemic *Echinacea paradoxa* (Norton) Britton.

Taken together, the study areas of this project represent a diversity of *Penstemon* habitats across the tallgrass prairie biome. Although we have pooled the floral forager lists in this paper (Table 1) by *Penstemon* taxa to conserve space, we emphasize the value of repeated samples of the same study areas over several years in evaluating the fidelity of floral foragers to specific penstemons.

The sites themselves are, for the most part, well-known nature preserves in Illinois, Kansas, and Missouri, for which there is a large literature available (e.g., Evers, 1955; Freeman & Hulbert, 1985; Ochs, 1993; Toney, undated; Solecki et al., 1986). These references contain much information on site history, location, ecology, and floristic composition.

**Floral fragrance.** Whole flowers of each species were placed in clean, glass, stoppered vials for periods up to two hours, following Buchmann et al. (1978). At the end of the two-hour period, the vials were uncorked and smelled.

**Bagging experiments.** To determine the potential role of self-compatibility and mechanical self-pollination (autogamy) in the absence of insect visitation, inflorescences were bagged during the flowering season. Nylon stockings or wood and mesh exclusion cages were placed over ten flowering shoots, in bud, of *P. cobaea* var. *cobaea*, twelve of *P. cobaea* var. *purpureus*, six of *P. digitalis* and three of *P. grandiflorus* for the length of individual flowering periods. Inflorescences were examined for the production of fruit and seed within two months following the withering of the last flower on the shoot.

**Pollinator analyses.** Field observations recorded foraging behavior of insects on, and within, *Penstemon* flowers. This included noting when foragers collected nectar and/or pollen and whether insects contacted anthers and stigmas while foraging.

The collection and analyses of floral foragers on *Penstemon* flowers represent the harvest of 35–50 insects/site each year. As these *Penstemon* populations represent protected species on nature pre-

serves, overcollection of prospective pollinators may lower seedset. Otherwise, the protocol for netting, killing, and processing specimens followed Bernhardt (1990a, b) and Bernhardt and Weston (1996). Floral foragers were collected on penstemon blossoms and killed in jars poisoned with ethyl acetate.

To verify the presence of *Penstemon* pollen, each euthanized insect was placed on a separate, clean glass slide and bathed in a few drops of absolute ethanol. At this time additional masses of pollen packed onto scopae and corbiculae were teased or scraped off with a dissecting needle. When the ethanol had evaporated, the pollen film left on the glass slide was stained with two to three drops of Calberla's fluid (Ogden et al., 1974). Because insects were sacrificed in a communal killing jar, there is the danger of pollen contamination as bodies of different insects contact each other (see Bernhardt & Weston, 1996). Therefore, we excluded pollen counts of less than 50 grains on insects since the vast majority of floral foragers were extremely hairy and liable to pick up loose grains due to static electricity. Pollen loads were counted on each slide as follows: a total of 200 pollen grains were counted for each insect in increments of five. If there were more than 150 grains of *Penstemon* pollen in the count (> 75%), the load was classified as a "*Penstemon* Pollen Only Load." If more than 150 grains of non-*penstemon* pollen was present the load was called an "Other Species Only Load." If more than 50 grains of both *Penstemon* and at least one other non-*Penstemon* species were present, this was called a "*Penstemon* + Other Species Load." If less than 200 grains were present on the slide, the load was scored as a "No Pollen Load."

Insect length was recorded by measuring six pinned specimens of each species, gender, and/or caste. The body length of each specimen was measured from the labrum to the apex of the abdomen (Bernhardt & Weston, 1996).

Insect specimens were identified by C. D. Michener, R. Brooks, and colleagues at Snow Entomological Museum, University of Kansas, and by M. Arduser of the Missouri Department of Conservation. Vouchers are deposited at the Snow Museum. Bee genera follow Michener et al. (1994).

## RESULTS

**Floral phenology.** Midwestern *Penstemon* spp. are found in bloom from early May until early July (Fig. 1). Flowering seasons overlap broadly between species. *Penstemon pallidus* is the first species to bloom in spring, while the two varieties of *P. cobaea*

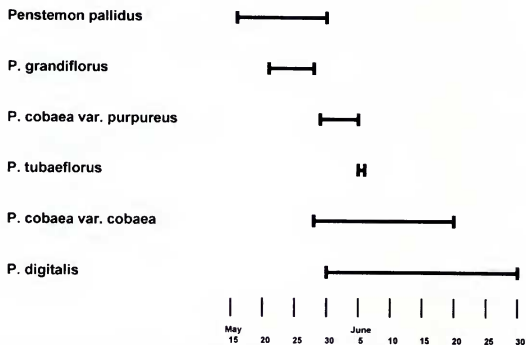


Figure 1. Floral phenology of *Penstemon* spp. (1994-1997).

are usually the last to begin flowering. *Penstemon digitalis* has the longest flowering period.

**Bagging experiments.** In both varieties of *P. cobaea*, no fruits were produced in the absence of visiting insects. In *P. grandiflorus* approximately 50% of the flowers produced full-sized capsules totally devoid of seed (parthenocarpy). In *P. digitalis*, the vast majority of flowers produced seed-containing capsules.

**Floral presentation.** All *Penstemon* spp. are protandrous with the stigma located within 2 mm of the two pairs of fertile anthers. All *Penstemon* spp. present their flowers horizontally with respect to the peduncle.

Flowers of all *Penstemon* spp. produce a barely discernible fragrance reminiscent of ripe cantaloupe. *Penstemon tubaeiflorus* is the only species studied with flowers that are pure, translucent-shiny white to the human eye. Flowers of *P. grandiflorus* appear pink-mauve with faintly discernible nectar guides. *Penstemon digitalis* and *P. pallidus* have white corollas with the floral throat streaked with narrow, light purple lines. *Penstemon cobaea* var. *cobaea* has pink or white corollas, while corollas of variety *purpureus* are deep purple (although we have records of red or white morphs in Taney Co., Missouri). Corollas of both varieties of *P. cobaea* have deep purple blotches and/or broad lines on the throat.

The corollas of *P. tubaeiflorus* (sinus 4-6 mm wide) have the most narrow floral tubes. The corollas of both varieties of *P. cobaea* (sinus 18-25 mm wide) and *P. grandiflorus* (15-18 mm wide) form large, bilabiate gullets. In contrast, the corol-

las of *P. digitalis* (sinus 8-12 mm) and *P. pallidus* (4-7 mm) form small gullets.

**Floral foragers.** Approximately 600 insects were caught foraging on *Penstemon* flowers. *Penstemon tubaeiflorus* was the only species that was visited consistently by the butterfly *Pierourus troilus*, but captures do not reflect the density of field observations (Table 1). The head and proboscis of this butterfly contacts stigmas and dehiscid anthers while it probes for nectar, with pollen loads deposited on the proximal portion of the proboscis (Table 2). Bees greater than 10 mm in length cling to the petal lobes while probing for nectar, acquiring dorsal depositions of pollen on their heads (Tables 1, 2).

In all remaining species pollination appeared to be dominated by Hymenoptera, with both varieties of *P. cobaea* also visited by bee flies (*Bombylius* spp.) that carried *Penstemon* pollen while contacting the stigmas (Table 2). Bumblebee flower-beetles (*Euphoria sepulchralis*, Scarabaeidae) collected on *P. cobaea* var. *cobaea* carried loads of *Penstemon* pollen, but they consumed basal floral organs and we were unable to determine whether they contacted stigmas (Table 2).

*Penstemon digitalis* and *P. cobaea* var. *purpureus* were visited by females of *Pseudomasaris occidentalis* (Vespididae). These wasps contacted the stigmas while swallowing pollen and/or foraging for nectar at the base of the corolla tubes, so pollen deposition was both ventral and dorsal.

The foraging behavior of bees was determined by physical size. Bees greater than 10 mm in length (*Anthophora*, *Bombus*, *Megachile*, and *Synhalonia*

Table 2. Size and behavior of floral foragers on *Penstemon* spp.

Floral forager	N = Number caught	$\bar{X}$ = Length of body* (mm)	Observed foraging for pollen	Observed foraging for nectar	Contacted anthers and stigma while foraging
<b>Coleoptera</b>					
<i>Euphoria sepulchralis</i>	6	12.5	-	+	-
<b>Diptera</b>					
<i>Bombylius</i> spp.	8	12.0	-	+	+
<b>Hymenoptera</b>					
<b>Bees</b>					
<i>Anthophora</i> spp. F	9	12.1	-	+	+
<i>Anthophora</i> spp. M	3	13.3	-	+	+
<i>Apis mellifera</i> W	1	13.0	-	+	+
<i>Augochlorella</i> spp. F	31	7.2	+	-	+
<i>Bombus</i> queens	118		-	+	+
<i>B. bimaculatus</i>	8	18.5			
<i>B. griseocollis</i>	5	22.8			
<i>B. nevadensis</i>	52	27.8			
<i>B. pennsylvanicus</i>	53	27.0			
<i>Bombus</i> workers	98		-	+	+
<i>B. bimaculatus</i>	43	15.3			
<i>B. fraternus</i>	10	—			
<i>B. griseocollis</i>	9	19.5			
<i>B. impatiens</i>	11	12.6			
<i>B. nevadensis</i>	9	18.7			
<i>B. pennsylvanicus</i>	16	22.5			
<i>Bombus</i> males					
<i>B. bimaculatus</i>	5	—	-	+	+
<i>Ceratina</i> spp. F	25	6.1	+	-	+
<i>Halictus ligatus</i> F	1	8.5	+	-	+
<i>Hoplitis pilosifrons</i> F	86	7.7	+	-	+
<i>Hoplitis pilosifrons</i> M	1	8.0	?	?	?
<i>Hoplitis producta</i> F	1	8.5	+	-	+
<i>Hylaeus</i> spp. F	4	5.0	+	-	+
<i>Lasioglossum</i>					
(= <i>Dialictus</i> ) spp. F	32	6.0	+	-	+
<i>Megachile brevis</i> M	1	11.0	-	+	+
<i>Osmia</i> spp. F	40	8.4	+	-	+
<i>Synhalonia hamata</i> F	40	15.3	-	+	+
<i>Synhalonia hamata</i> M	1	12.5	-	+	+
<i>Synhalonia rosae</i> F	1	14.2	-	+	+
<i>Synhalonia rosae</i> M	5	13.7	-	+	+
<i>Xylocopa virginica</i> F	4	21.4	?	?	?
<i>Xylocopa virginica</i> M	1	22.5	?	?	?
<b>Wasps</b>					
<i>Pseudomasaris</i> sp. F	3	19.0	+	+	+
<b>Lepidoptera</b>					
<i>Pterourus troilus troilus</i> F	4	22.5	-	+	+/?
<i>Pterourus troilus troilus</i> M	1	22.0	-	+	+/?

\* Measurement refers to body length measured from the clypeus (excluding the length of the proboscis) to the tip of the abdomen.

F = female, M = male, Q = queen, W = worker.

spp.) entered the floral tube and probed for nectar, receiving dorsal depositions of pollen from dehiscent anthers while contacting the stigmas (Table 2). These bees did not collect *Penstemon* pollen by scraping anthers with their legs. Although *Xylocopa virginica* was over 10 mm in length, bees of this species mostly obtained nectar from *Penstemon* flowers by piercing the base of the corolla tube without entering the floral throat. Observations of queen and worker *Bombus* spp. showed that these large bees typically foraged first on the lowest open flower on an inflorescence, and then climbed up the inflorescence following the spiral of open corollas and visiting flowers in both phases of protandry. All the large bee species were observed visiting the flowering shoots of several genets in succession.

In contrast, bees less than 10 mm in length (*Augochlorella*, *Ceratina*, *Hoplitis*, *Hylaeus*, *Lasioglossum*, and *Osmia* spp.) were not observed to probe for nectar. These bees were observed clinging upside-down and collecting pollen from dehiscent anthers. *Penstemon* pollen was deposited ventrally on these insects, and bees contacted stigmas while foraging for pollen. These small bees did not appear to discriminate between flowers in the young (dehiscent) versus old (empty) anther phases of protandry in *Penstemon* flowers based on corolla features. These bees were observed to enter the floral tube, cling to the old, empty anthers and contact the stigmas for a few seconds, but then they exited the flowers without attempting to scrape old anthers. These small bees were observed visiting several flowering shoots in succession before leaving the site.

Approximately 57% of all bees captured on *Penstemon* spp. carried pure loads of *Penstemon* pollen in their corbiculae or scopae (Table 1). An additional 20% carried *Penstemon* pollen mixed with the pollen of other co-flowering species. Mixed pollen loads included both nectar-producing (*Baptisia* spp., *Delphinium* spp., *Onosmodium* sp., *Pycnanthemum* sp., *Rubus* sp., and *Teucrium* sp.) and nectarless (*Rosa* spp., *Schrankia nuttallii*) species. Pollinaria of several *Asclepias* spp. were found attached exclusively to the first pair of legs.

Some queen bumblebees appeared to restrict their foraging to selected *Penstemon* spp. *Bombus pennsylvanicus* was most abundant on the large-gullet species: *P. cobaea* and *P. grandiflorus*. *Bombus nevadensis* was found primarily on the small, gullet-shaped corollas: *P. digitalis* and *P. pallidus*. No morphometric difference could be found between *B. pennsylvanicus* and *B. nevadensis* queens. Bumblebees were seldom observed or collected in flowers of populations of *P. cobaea* var. *cobaea* that con-

sisted of < 50 flowering shoots (Konza Prairie sites 1995 and 1997).

The density of bumblebees belonging to the neuter, worker caste was highly skewed in the data set. Ninety percent of all workers of six *Bombus* spp. (Table 1) were restricted to the flowers of *P. digitalis* at the Litzsinger Road Ecology Center, a restored site. The remaining 10% were distributed among all remaining study sites for all *Penstemon* spp. Although the flowering period of *P. digitalis* at the Litzsinger Road site overlapped broadly with *P. digitalis* at the two remaining sites, the presence of worker-caste bumblebees on virgin prairies and on old restored prairies was only a fraction of the total catch of workers of *Bombus* species at a five-year restoration site.

The density and diversity of bees less than 10 mm long differed among the bee-pollinated *Penstemon* spp. (Table 1). Over 50% of all bees collected on *P. cobaea* var. *cobaea* were *Hoplitis pilosifrons* (Megachilidae). In contrast, approximately 45% of bees on *P. grandiflorus* were *Augochlorella striata* (Halictidae). Most of the small-bodied bees collected on *P. digitalis* were *Lasioglossum* spp. (Halictidae). *Osmia* spp. (Megachilidae) and *Hoplitis pilosifrons* were equally represented on *P. pallidus*. Note that most of these bee taxa were collected in smaller proportions on *Penstemon* flowers other than the modal *Penstemon* species (listed above) at each site (Table 1). These smaller bees were observed to visit more than one open flower on an inflorescence, and to visit several flowering shoots in succession. Small bees were rarely observed and collected on flowers of *P. cobaea* var. *cobaea* in populations of > 500 flowering shoots (Konza Prairie sites 1995 and 1997). These results on the foraging preferences of bumblebees versus small, solitary bees on *P. cobaea* populations were highly significant by chi square ( $T = 37.33$ ,  $P \ll 0.001$ ). Statistical procedures follow Conover (1980). Remembering that the square root of the chi square test statistic is approximately the absolute value of the test statistic of the normal distribution, we can estimate that there is an extremely low probability, on the order of one occurrence in a million, that these results are due to chance alone (Stanley Sawyer, pers. comm.). We also emphasize that we do not have a high enough level of sample replication in our data set to use parametric statistics, which would distinguish species effects from site effects, and acknowledge that we thus cannot make definitive comments here on this issue.

#### DISCUSSION

Pollen dispersal varies among prairie *Penstemon* species in the American Midwest. The putative



trend toward butterfly pollination in *Penstemon tubaeiflorus* is reflected by the constriction of the floral tube. Otherwise, there appeared to be few floral characters indicative of classic psychophily in this species (Barth, 1985; Proctor et al., 1996). For example, flowers were not held erect and lacked the characteristic pigmentation associated with "butterfly flowers." This suggests that this pollination system is recent and that this species may be derived from a bee-pollinated ancestor.

In the four remaining *Penstemon* spp., bee visitation was dominant, involving a broad diversity of potential pollinators representing four families of Apoidea. Furthermore, bee pollination in mid-western *Penstemon* spp. appeared to be a two-tiered system. Pollination by bees in most angiosperm systems is based on either passive or active contact (sensu Bernhardt, 1996) with the anthers. Pollination by passive contact implies that the bee first contacts dehiscent anthers and receptive stigmas while foraging for nectar, or other rewards, toward the base of the flower. Pollination by active contact suggests that the bee contacted the receptive stigma while foraging specifically for pollen. Both modes of pollination occurred in four *Penstemon* spp., but each pollination mode was performed by two different sets of bees. Large-bodied, long-tongued bees effected passive pollination, while small-bodied bees, with tongues of varying lengths, effected active pollination. Since *Penstemon* flowers were protandrous, this implies that active pollination required automimicry (sensu Dafni, 1993) to encourage cross-pollination. Small bees foraging for pollen were unable to discriminate initially between fresh, dehiscent anthers and old, empty anthers, suggesting that pollen collection in *Penstemon* flowers represents a form of partial reinforcement for smaller bees.

We have found no previous citations of pollination of *Penstemon* spp. by bumblebee flower-beetles (*Euphoria sepulchralis*). These greenish scarabs are not hairy, but became dusted with *Penstemon* pollen in the process of feeding on nectar and floral organs while residing inside the base of the corolla tube for long periods of time. It is reasonable to suggest that they played a minor role in the pollination of *Penstemon* spp. on the Konza Prairie Research Natural Area. Likewise, uncommon pseudomasarid wasps probably contributed to the pollination of *P. digitalis* and *P. cobaea* var. *purpureus*. The literature suggests that pseudomasarids are important pollinators of some *Penstemon* spp. native to the west coast of North America (Straw, 1956).

Three patterns of interactions between *Penstemon* flowers and bees were documented. First, the

foraging specificity of queens of *Bombus nevadensis* to *P. digitalis* and *P. pallidus* versus that of *B. pennsylvanicus* to *P. cobaea* and *P. grandiflorus* provides an interesting dichotomy. The morphometric differences between the two *Bombus* spp. with respect to glossa length and body parameters are minor, according to taxonomic authorities (Medler, 1962). In both species, the combined length of the glossa and prementum is about 12.5 mm. Based on our collections, it would appear that *B. nevadensis* preferred *Penstemon* corollas with small gullets, while *B. pennsylvanicus* preferred corollas with large gullets and much expanded sinuses. (In contrast, both *B. griseocollis* queens, which rarely visit penstemons, and *B. fraternus* queens, which did not visit penstemons at all in this study, have a combined glossa plus prementum length of about 10 mm.) However, statistical analyses of the foraging preferences of queens of *B. nevadensis* and *B. pennsylvanicus* would require additional data sets from a far greater number of sites. In fact, we acknowledge that the above-mentioned relationship between certain *Bombus* queens and *Penstemon* spp. may actually reflect a combination of overlapping phenologies and biogeographies.

Second, bumblebee queens far outnumbered neuter workers on *Penstemon* flowers. This was to be anticipated considering the well known, annual life-cycle of *Bombus* spp. in the Northern Hemisphere, since *Bombus* queens always forage for pollen in spring until they raise a significant retinue of workers (Heinrich, 1979). What was not expected was the relative abundance of workers on restoration sites ( $N = 99$ ) versus the relative absence of workers ( $N = 7$ ) on both original, tallgrass prairies and true glade sites. We were unable to determine whether *Bombus* workers simply avoided *Penstemon* populations in "virgin" prairies and glades or whether *Bombus* colonies matured more rapidly within, or adjacent to, restored sites. Whatever the case, this pattern, if continuous, may have direct implications for future conservationists who emphasize the restoration of original pollination systems.

Third, collections of small-bodied, solitary bees indicated selective foraging preferences that correlated negatively with the size of *Penstemon* populations. This occurred exclusively in *P. cobaea* var. *cobaea* within sites on the Konza Prairie Research Natural Area. Over four seasons, 59 solitary bees were collected on populations with less than 50 flowering stems, but only four specimens were ever collected on populations with more than 500 flowering stems. These massively flowering populations of *P. cobaea* appeared to be pollinated primarily by *Bombus* queens. Why did solitary bees avoid the

greater resources offered by a much higher density of *Penstemon* flowers? Our only explanation is that *Bombus* queens outcompeted solitary bees for access to *Penstemon* corollas in larger populations, even though they did not compete for the same resource. Bernhardt (1993) mentioned older reports that female xylocopines (Anthophoridae) recognized and avoided flowers of *Passiflora* spp. that had been visited and marked with a glandular secretion by another female of the same species. Perhaps solitary bees recognized and avoided *Penstemon* flowers that had been visited and marked by *Bombus* spp. The comparative disinterest that *Bombus* queens showed to smaller populations of *P. cobaea* may have reflected both economic and energetic considerations. As bumblebees are strong fliers and trapline foragers (Heinrich, 1979; Bernhardt & Montalvo, 1979) they may have avoided smaller populations of flowering plants because such populations offered weaker visual/olfactory displays during peak flowering periods. While a traplining forager would seem most appropriate for the pollination of smaller, isolated populations, their visitation contradicts bumblebee economics (Heinrich, 1979). Small flowering populations provide inadequate rewards for both the bees and the larvae provisioned by the foraging queens (Bernhardt, 1990b).

Waser et al. (1996) argued that generalized pollination is the rule, not the exception, in most angiosperm species. Floral generalization becomes selectively advantageous when pollinator populations fluctuate, so that pollinator shifts can occur in angiosperm populations.

We do not accept that bee pollination in all mid-western *Penstemon* spp. must also reflect fluctuations in generalized trends favoring either large or small bees. We interpret bee pollination in four *Penstemon* spp. as two specialized syndromes concurrent within the same flowers. A coexisting, two-tiered syndrome has been selected for pollination by both large-bodied, long-tongued, nectar-foraging bees and small-bodied, pollen foragers. Note that all *Bombus* spp. and most of the solitary bees (excluding *Osmia* s. str.) collected in this study are polylectic taxa in grasslands (Bernhardt, 1990a, b). Note also that the same bee species may adopt either active or passive pollen collection on different, coblooming flowers (Bernhardt, 1996).

With two bee-pollination syndromes operating in the same *Penstemon* flower, either syndrome can encourage outcrossing, regardless of demographics, in any *Penstemon* population. It seems naive to presume that dichotomous modes of pollination in the same flower usually represent shifts in generalized

patterns. For example, the older literature has long suggested that a two-tiered system has existed in some species pollinated by both birds and bees. Both birds and bees contribute to pollination in one species because two different modes of attractants and rewards overlap within the same flower (Grant & Grant, 1968; Breedlove, 1969; Macior, 1975; Grant, 1976; Schemske, 1978). If bees and homeothermic vertebrates can pollinate the same flower, we should also be able to postulate two different groups of bees pollinating the same flower for different rewards. Frankly, a generalized mode of pollination must always be implied when field studies fail to incorporate basic analyses of pollen loads and observations of foragers contacting receptive stigmas.

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